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Insular and Anaplastic Carcinoma of the Thyroid

A 45-Year Comparative Study at a Single Institution and a Review of the Significance of p53 and p21

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Objective
To analyze the clinicopathologic features of a large cohort of patients with insular or anaplastic carcinomas treated at a single institution.

Summary Background Data
Insular and anaplastic carcinomas of the thyroid, although uncommon, have more aggressive clinical behavior than well-differentiated carcinomas of the thyroid. In the literature, the incidence and features of these carcinomas have not been fully characterized.

Methods
The authors reclassified 740 primary thyroid carcinomas diagnosed and treated between January 1, 1954, and December 30, 1998, to select those with features that met the histologic criteria of insular or anaplastic carcinoma. The clinicopathologic features of these carcinomas were studied and compared. The expression of p53 and p21 in these tumors was analyzed by immunohistochemistry.

Results
Twenty-two patients (5 men, 17 women) with insular carcinoma and 38 patients (7 men, 31 women) with anaplastic carcinoma were found. Patients with insular carcinomas were younger (mean age 45 vs. 70 years) and had smaller tumors than those with anaplastic carcinomas (mean diameter 5 vs. 8 cm). Insular carcinomas were commonly mislabeled as other histologic subtypes, whereas anaplastic carcinomas might be overdiagnosed on pathologic examination. A history of long-standing goiter (>10 years) was noted in 27% of patients with insular carcinoma and 24% of patients with anaplastic carcinomas. Concomitant well-differentiated carcinomas of the thyroid were noted in 59% of patients with insular carcinoma and 39% of patients with anaplastic carcinoma. In anaplastic carcinomas, 13% of patients had concomitant insular carcinoma. Calcification or bone was noted in the stroma of 23% of patients with insular carcinomas and 47% of those with anaplastic carcinomas. The 10-year survival rates for patients with insular carcinoma and anaplastic carcinoma were 42% and 3%, respectively. Distant metastases were seen in 32% of patients with insular carcinoma and in 47% of patients with anaplastic carcinomas. In both types of carcinomas, metastatic tumors were often seen in bone and lung. Distant metastases were noted in a variety of organs in anaplastic carcinomas. In insular carcinoma, neither p53 nor p21 expression was present. In anaplastic carcinoma, p53 and p21 expression was identified in 69% and 3%, respectively. Concomitant expression of p53 and p21 was noted in one tumor.

Conclusions
Insular carcinoma and anaplastic carcinoma had distinctive clinicopathologic features, and recognition of these histologic variants is important for better management of these tumors in the future. p53 overexpression might have a role in dedifferentiation from insular carcinoma to anaplastic carcinoma.

Carcangiu et al[1] characterized a new entity of thyroid carcinoma termed insular carcinoma (also called poorly differentiated carcinoma) in 1984. The tumor was situated morphologically and biologically in an intermediate position between the well-differentiated (papillary and follicular) and the anaplastic thyroid carcinomas. The histologic features of the insular carcinomas include formation of solid clusters (insulae) of tumor cells containing a variable number of small follicles; variable but consistently present mitotic activity; capsular and blood vessel invasion; and frequent necrotic foci, sometimes leading to formation of peritheliomatous patterns. To date, more than 150 patients with insular carcinoma have been described in the literature.[1–8] However, most studies of this entity were case

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reports rather than case series. In addition, many authors used the term "poorly differentiated thyroid carcinoma" to mean well-differentiated thyroid carcinoma that exhibited a focal solid, trabecular or scirrhous pattern[9] instead of what was originally characterized by Carcangiu et al.[1] Also, many authors used the term without definition. Thus, it is difficult to characterize the clinicopathologic features and incidence of this subtype of thyroid carcinoma.

Mutation or deletion of the p53 gene is one of the most frequently detected genetic changes in human cancers.[10] p21 protein has been described as the critical downstream effector in the p53-specific pathway of growth control and can also be induced by an alternative, p53-independent pathway.[11,12] The genes have been proposed to play important roles in thyroid carcinomas.[13–40]

To determine the characteristic features of insular and anaplastic (undifferentiated) carcinomas, we reclassified all the primary thyroid tumors reported during the past 45 years in our hospital to select those that met the histologic criteria of insular or anaplastic carcinoma. The potential roles of p53 and p21 expression in these tumors were also investigated.

METHODS

Data Collection

Histologic results of 740 primary thyroid carcinomas reported between January 1, 1954, and December 31, 1998, were reviewed. The tumors with features of anaplastic or insular carcinomas were selected for further investigation. They were classified according to the presence of a predominant component with the most aggressive behavior. We observed strictly the criteria as originally proposed by Carcangiu et al[1] when labeling a tumor as insular carcinoma (Fig. 1) and the criteria of the Armed Forces Institute of Pathology[41] when labeling a tumor as anaplastic carcinoma. Immunohistochemical studies (using antibodies against thyroglobulin, cytokeratins, leukocyte common antigen, L26, CD3, calcitonin, and others) were performed when necessary to confirm the diagnoses.

The patients included were all ethnic Chinese. Sixty patients with insular (n = 22) or anaplastic carcinomas (n = 38) of the thyroid were found. There were 12 men and 48 women, with ages ranging from 15 to 93 years (median age 64 years). The clinical presentation, preoperative clinical diagnosis, pathologic diagnosis, treatment, and outcome of the patients were examined. On pathologic examination, the gross appearance, location, presence of concomitant well-differentiated thyroid carcinoma, histologic patterns, and stromal calcification or ossification of these tumors were noted. Postmortem examination was performed in 12 patients (2 with insular carcinomas and 10 with anaplastic carcinomas).

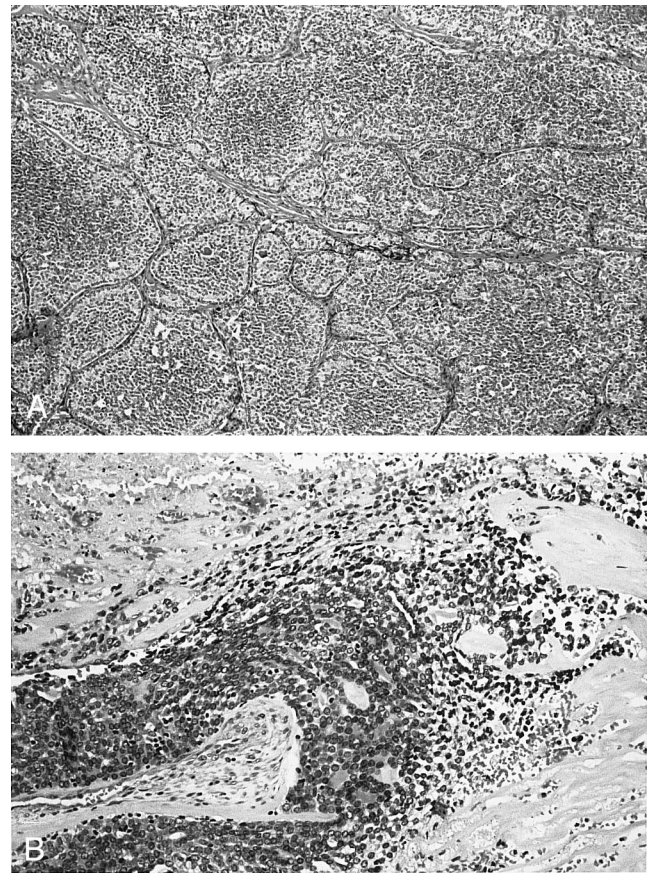

Figure 1. Histologic features of insular carcinoma. (A) Insular pattern (hematoxylin and eosin, ×150). (B) Peritheliomatous pattern (hematoxylin and eosin, ×200).

Immunostaining for p53 and p21

Representative paraffin blocks from the thyroid carcinomas were obtained to study the expression of p53 and p21. The block was considered to be adequate for the study only if it was of adequate size (surface area >4 cm^2 and $>50\%$ of the surface area occupied by the tumor). Thirteen insular and 29 anaplastic carcinomas met the criteria and were then selected for the analysis of p53 and p21 expression.

From the selected paraffin blocks, 5-μm-thick sections were cut. They were deparaffinized with xylene and rehydrated through graded concentrations of alcohol. The sections were then treated with microwave irradiation at 95°C in 10 mmol/L citrate buffer for 9 minutes. They were washed in water, rinsed with Tris-buffered saline (TBS), and treated with 3% hydrogen peroxide in methanol for 10 minutes at room temperature to block the intrinsic peroxidase activity. The sections were again washed with water and rinsed with TBS. Ten percent normal rabbit serum was then added at room temperature for 10 minutes. Afterward, primary mouse monoclonal anti-p53 antibody (NCL-p53-DO7 at dilution 1:100) (Novocastra Laboratories Ltd., Newcastle upon Tyne, UK) and anti-p21 antibody at dilution 1:40 (Novocastra) were added and incubated in a moist

Table 1. CLINICOPATHOLOGIC FEATURES OF INSULAR CARCINOMA OF THE THYROID

Patient	Sex/ Age	Clinical Diagnosis	Original Pathologic Diagnosis	Preexisting Long-Standing Goiter	Size (cm)	Site	Co. WDCA	Cal.	Site(s) of Met.	Follow-Up
1	F/53	Carcinoma	Unclassified carcinoma	No	2.8	Left	No	Yes	Bone	—
2	F/54	Carcinoma	Unclassified carcinoma	Yes, >20 years	5.5	Right	No	Yes	—	—
3	F/63	Carcinoma	Unclassified carcinoma	No	5.0	Left	Yes	No	Lung, heart	Died within 1 mo
4	F/26	NH	Unclassified carcinoma	No	4.0	Right	Yes	No	—	Alive after 39 y
5	M/54	NH	Unclassified carcinoma	No	4.5	Right	No	No	Lung	Died after 1 y
6	F/15	Carcinoma	Insular carcinoma	No	2.5	Left	No	No	—	Alive after 28 y
7	F/28	NH	Follicular carcinoma	No	2.0	Left	No	No	—	—
8	F/62	NH	Follicular carcinoma	Yes, >30 years	12.0	Left	Yes	No	—	—
9	M/54	Carcinoma	Follicular carcinoma	No	3.5	Right	Yes	No	Bone	Alive with disease after 2 y
10	M/62	Carcinoma	Insular carcinoma	No	5.0	Right	No	No	Liver	Died after 9 m
11	M/75	Carcinoma	Follicular carcinoma	No	6.0	Left	Yes	No	—	Alive after 16 y
12	F/47	Carcinoma	Follicular carcinoma	No	2.5	Right	Yes	No	—	Alive after 22 y
13	F/64	Carcinoma	Follicular carcinoma	Yes, >20 years	9.5	Left	No	No	—	—
14	F/63	Carcinoma	Papillary carcinoma	No	5.5	Right	No	No	—	—
15	M/79	Carcinoma	Insular carcinoma	No	11.0	Isthmus	No	No	—	Died within 1 m
16	F/25	Carcinoma	Follicular carcinoma	No	2.0	Left	No	No	—	Alive after 17 y
17	F/64	Carcinoma	Follicular carcinoma	Yes, >20 years	6.0	Left	Yes	Yes	Lung	Died after 13 y
18	F/67	Carcinoma	Follicular carcinoma	Yes, >35 years	7.0	Left	Yes	Yes	Bone	Died after 4 y
19	F/64	Carcinoma	Follicular carcinoma	No	1.1	Left	Yes	Yes	Bone	Died after 9 y
20	F/55	Carcinoma	Anaplastic carcinoma	Yes, >10 years	4.5	Right	Yes	No	—	Alive after 5 y
21	F/39	Carcinoma	Follicular carcinoma	No	4.0	Left	Yes	No	—	Alive after 3.5 y
22	F/73	Carcinoma	Anaplastic carcinoma	No	5.5	Left	No	No	—	Alive after 6 m

Cal, presence of calcification or osseous metaplasia; Co. WDCA, concomitant well-differentiated thyroid carcinoma; Met., metastasis(es); m, month(s); NH, nodular hyperplasia; y, year(s).

chamber overnight at 4°C. The slides were again washed three times in TBS for 3 minutes. Rabbit antimouse biotinylated IgG (diluted in 10% normal rabbit serum, E354) (Dako, Gloustrup, Denmark) and preincubated (30 minutes at room temperature) avidin–biotin complex (1:100) (Amersham, Buckinghamshire, UK) were added for 30 minutes at 37°C. They were washed in TBS as before and were then developed in freshly prepared DAB/H_2O_2 solution for 10 minutes at room temperature. The sections were then washed in water, counterstained with Mayer's hematoxylin for 1 minute at room temperature, dehydrated, cleaned, and mounted. Negative controls were sections treated the same as above but with omission of the primary antibodies. Paraffin blocks of esophageal squamous cell carcinoma known to be strongly positive for p53 protein and p21 protein were used as the positive controls. Brown nuclear stain was regarded as positive.

Statistical Analysis

Statistical analysis was performed using the Student *t* test with Yates correction (continuous variables) and Fisher exact test (categorical variables). The actuarial survival rate of the patients was calculated from the date of presentation to the date of death or last follow-up using the Kaplan-Meier method. The potential impact of various factors on survival was studied by the log-rank test. Significance level was taken at $P < .05$.

RESULTS

Insular Carcinomas

There were 22 patients (5 men, 17 women) with insular carcinoma of the thyroid (Table 1). The mean age of these patients was 54 years (range 15–79). They accounted for 3% of the primary thyroid carcinomas. At presentation, all except one had a neck mass. The remaining patient had bony metastasis before the detection of the primary thyroid tumor. A malignant thyroid tumor was diagnosed clinically in 18 patients (the other 4 were diagnosed as having nodular hyperplasia). A history of long-standing goiter lasting for more than 10 years was present in 27% (6/22) of patients at presentation. Treatment included surgery (n = 12), surgery plus radiotherapy (n = 8), and radiotherapy (n = 1). One patient died before commencement of treatment.

Thirteen tumors were in the left lobe, eight in the right lobe, and one in the isthmus of the thyroid. The mean diameter of these lesions was 5 cm (range 1.1–12). They were diagnosed initially as follicular carcinomas (n = 11), unclassified carcinomas (n = 5), poorly differentiated carcinomas (n = 3), anaplastic carcinomas (n = 2), and pap-

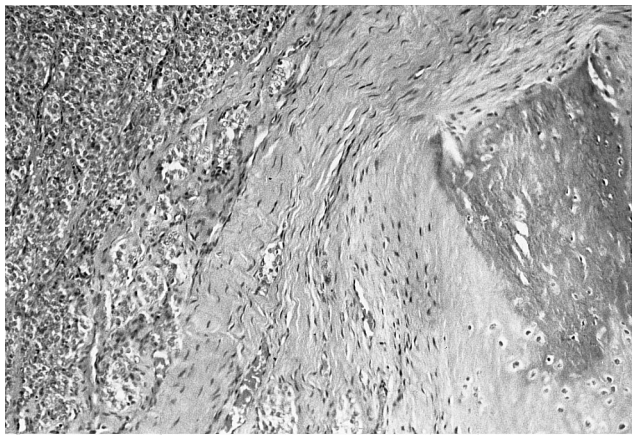

Figure 2. Insular carcinoma of thyroid with osseous metaplasia in the stroma (hematoxylin and eosin, ×150).

illary carcinoma (n = 1). Concomitant well-differentiated thyroid carcinomas were identified in 59% (13/22) of the tumors; calcification or bone formation was seen in the tumor stroma in 23% (n = 5) (Fig. 2). On follow-up, distant metastases were seen in seven patients. Metastatic tumors were found in bones (n = 4), lungs (n = 3), heart (n = 1), and liver (n = 1).

Anaplastic Carcinoma

There were 38 patients (7 men, 31 women) with a mean age of 70 years (range 15–93) (Table 2). They accounted for 5% of the primary thyroid carcinomas. All the patients had a rapid enlarging neck mass noted at presentation. The clinical diagnoses were thyroid cancer (n = 35) or cancers of the upper aerodigestive tract (n = 3). A history of long-standing goiter (>10 years) was present in 24% (9/38) of patients. The patients were managed by surgery alone (n = 11), radiotherapy alone (n = 3), surgery plus radiotherapy (n = 9), or surgery plus radiotherapy and chemotherapy (n = 3). The remaining 12 patients died before commencement of therapy.

Seventeen tumors (45%) were in the left lobe, 15 (39%) in the right lobe, and 2 (5%) in the isthmus of the thyroid, and 4 (11%) showed diffuse infiltration in both lobes. The mean diameter of these lesions was 8 cm (range 3–25). All 38 tumors had been labeled as anaplastic carcinoma by the original pathologists. Eight tumors originally labeled as anaplastic carcinomas were rejected after histologic review and further immunohistochemical studies. These eight were in fact lymphomas (n = 6) or insular carcinomas (n = 2). Concomitant well-differentiated thyroid carcinoma and insular carcinoma of the thyroid were present in 39% (n = 15) and 13% (n = 5) of the tumors, respectively (Fig. 3). In two patients, well-differentiated, insular and anaplastic carcinomas were present. One of these patients had insular carcinoma in the thyroid but foci of anaplastic carcinoma in the lymph nodes. Four patients had previous resection of papillary carcinoma (at 2, 4, 5.5, and 22 years) before the occurrence of anaplastic carcinoma.

Twenty-four anaplastic carcinomas had a predominately spindle cell pattern. One tumor had a hemangiopericytoma-like pattern. The others had either a predominately giant cell (18%; n = 7) or epidermoid (18%; n = 7) pattern (Fig. 3). Calcification or bone formation was seen in the tumor stroma in 47% (n = 18) of the tumors. Distant metastases were seen in 18 patients during follow-up or after death. Metastatic carcinomas were found in lungs (n = 14), bones (n = 5), heart (n = 4), liver (n = 3), kidneys (n = 2), adrenals (n = 2), brain (n = 1), pancreas (n = 1), and salivary glands (n = 1) in these patients.

p53 and p21 Expression

p53 and p21 staining was negative in insular carcinomas. p53 overexpression was seen in 69% (20/29) of the anaplastic carcinomas (Fig. 4). In patients with concomitant well-differentiated or insular carcinomas, the p53 positivity was noted only in the anaplastic component. Focal p21 staining was noted in one patient (Fig. 5). The difference of p53 expression between insular and anaplastic carcinoma was highly significant ($P = .0001$). Factors including age, gender, history of long-standing goiter, and size and histologic patterns of the tumors did not contribute significantly to the difference in p53 overexpression among the anaplastic carcinomas.

Comparison of Survival Time

Follow-up data were available on 16 patients with insular carcinomas and 34 patients with anaplastic carcinomas. During a mean follow-up period of 10 years, 9 of 16 patients with insular carcinomas were alive and disease-free. The median survival was 13 years. The overall 5- and 10-year survival rates were 46% and 42%, respectively (Fig. 6). In contrast, during a mean follow-up period of 2 years, all but two patients with anaplastic carcinomas died. The median survival was 2 months. The overall 5- and 10-year survival rates were 15% and 3%, respectively. Thus, the survival of the two groups of patients was significantly different ($P = .0001$). Also, patients 60 years or older had a median survival of 2 months, compared with 56 months in younger patients ($P = .0019$). Patients with a tumor 6 cm or larger had a shorter survival than patients with a smaller tumor (2 months vs. 13 months, $P = .003$). In addition, patients with tumors with p53 overexpression had worse survival than those without ($P = .01$).

DISCUSSION

Insular carcinoma of the thyroid is an uncommon pathologic entity, and many of the tumors have been documented as case reports in the literature.[1–8] Thus, the exact incidence of insular carcinoma remains unknown. Insular carcinoma

Table 2. CLINICOPATHOLOGIC FEATURES OF ANAPLASTIC CARCINOMA OF THE THYROID

Patient	Sex/ Age	Preexiting Long-Standing Goiter	Size (cm)	Site	*p53*	p21	Co. WDCA	Co. Insular Carcinoma	Histologic Pattern	Cal.	Follow-Up	Site(s) of Metastasis(es)
1	M/15	No	4.0	Left	−	−	No	No	Spindle cell	No	Died after 5 m	Bone
2	F/71	No	10.0	Right	ND	ND	No	No	Spindle cell	No	Died after 1 m	—
3	F/53	No	7.5	Right	ND	ND	No	Yes	Giant cell	No	Died after 6 m	Lung, heart
4	M/87	No	8.0	Right	ND	ND	No	No	Spindle cell	Yes	Died 1 d after admission	Lung, bone, heart, kidney
5	F/70	Yes, >20 years	9.0	Left	ND	ND	No	No	Spindle cell	No	Died 2 d after admission	Heart, liver
6	F/84	No	10.0	Left	+	−	No	No	Spindle cell	No	Died 11 d after admission	Lung, liver
7	F/52	No	4.0	Right	+	−	No	No	Giant cell	No	—	—
8	F/60	Yes, >20 years	9.0	Right	−	−	No	No	Spindle cell	No	Died 12 d after admission	Lung, adrenal gland, brain
9	F/47	No	4.0	Left	−	−	No	Yes	Giant cell	No	Died after 56 m	Lung, liver, pancreas
10	F/86	Yes, >30 years	25.0	Left	+	−	No	No	Spindle cell	No	Died 7 d after admission	—
11	F/71	No	10.0	Diffuse	+	−	Yes	Yes	Spindle cell	No	—	Lung
12	F/75	Yes, >40 years	13.0	Right	+	−	Yes	No	Epidermoid	No	—	—
13	M/64	No	7.0	Right	+	−	No	No	Epidermoid	No	Alive after 17 y	—
14	M/82	No	10.0	Diffuse	ND	ND	Yes	No	Spindle cell	Yes	Died after 1 m	Lung, adrenal gland, heart, kidney
15	F/63	Yes, >20 years	15.0	Left	+	−	Yes	No	Spindle cell	Yes	Died after 6 m	Parotid gland, submandibular gland
16	F/93	No	5.0	Right	ND	ND	No	No	Spindle cell	Yes	Died on day of admission	—
17	F/74	No	12.0	Right	+	−	No	No	Giant cell	Yes	—	—
18	F/88	Yes, >10 years	10.0	Left	+	−	No	No	Spindle cell	No	Died after 41 d	—
19	F/64	No	6.0	Isthmus	−	−	Yes	No	Epidermoid	Yes	Died after 14 m	—
20	F/80	No	8.0	Left	−	−	Yes	No	Epidermoid	Yes	Died after 4 m	—
21	F/80	No	15.0	Diffuse	+	−	No	No	Spindle cell	No	Died after 3 d	Lung
22	M/55	No	7.0	Right	+	−	Yes	No	Spindle cell	No	Died after 5 m	—
23	F/80	No	5.0	Left	+	−	Yes	No	Epidermoid	No	Died after 1 m	—
24	M/55	No	5.5	Right	+	−	Yes	No	Spindle cell	No	Died after 10 m	Bone
25	F/54	No	8.0	Left	+	+	No	No	Spindle cell	Yes	Died after 5 m	Bone
26	F/63	No	10.0	Left	+	−	Yes	No	Spindle cell	No	Died after 7 m	—
27	F/76	No	7.0	Right	+	−	No	No	Spindle cell	Yes	Died after 2 m	Lung
28	F/84	Yes, >20 years	10.0	Left	+	−	Yes	No	Epidermoid	Yes	Died after 1 m	Lung
29	F/68	No	7.5	Right	−	−	Yes	No	Spindle cell	No	Died after 2 m	—
30	F/65	No	10.0	Right	−	−	No	No	Giant cell	No	Died after 10 m	—
31	F/86	No	10.0	Left	+	−	Yes	No	Epidermoid	No	Died after 8 m	—
32	F/72	No	3.0	Left	−	−	No	No	Spindle cell	No	Alive after 73 m	—
33	F/82	No	6.0	Right	+	−	Yes	No	Giant cell	Yes	Died after 10 m	Lung
34	F/69	No	6.0	Left	−	−	Yes	No	Spindle cell	No	Died after 11 m	Lung
35	F/69	Yes, >20 years	5.0	Right	+	−	No	No	Giant cell	Yes	Died after 9 m	Lung
36	F/66	Yes, >10 years	14.0	Diffuse	ND	ND	No	No	Spindle cell	No	Died after 15 d	—
37	M/73	No	3.8	Right	ND	ND	No	No	Spindle cell	No	Died after 5 m	Bone
38	F/87	No	4.0	Isthmus	ND	ND	No	No	Spindle cell	No	Died after 11 d	—

Cal, presence of calcification or osseous metaplasia; Co., concomitant; d, day(s); Met., metastasis(es); m, month(s), ND, not done; WDCA, well-differentiated thyroid carcinoma; y, year(s).

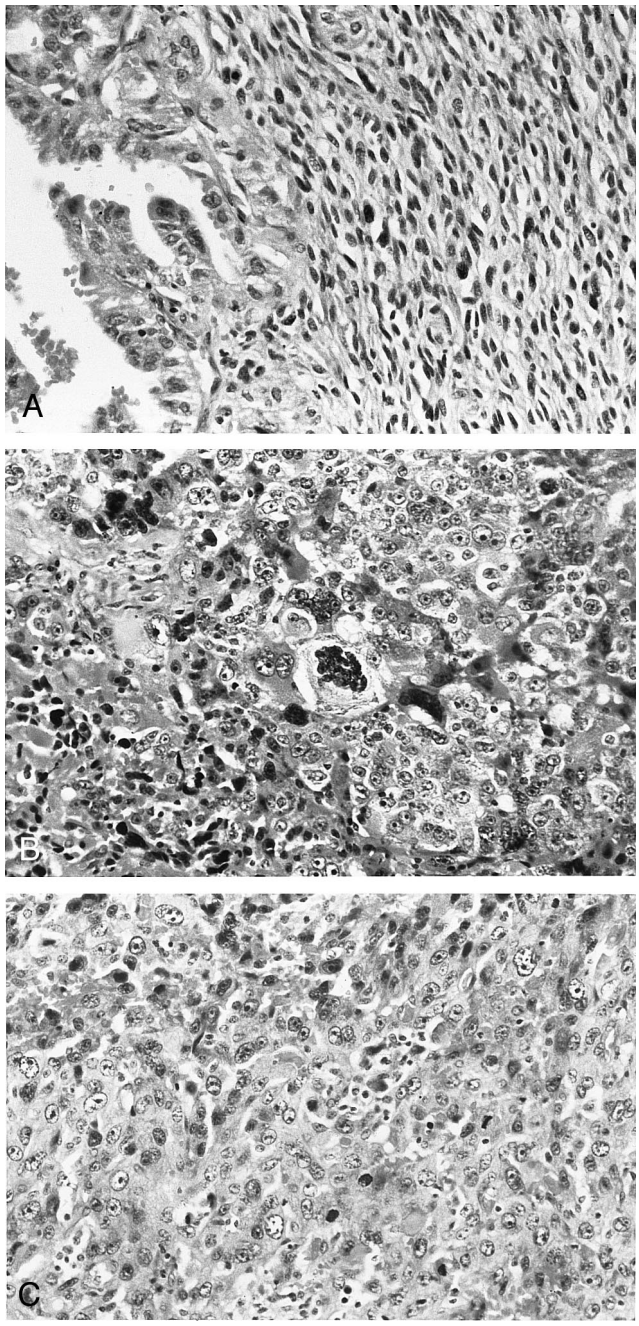

Figure 3. Anaplastic carcinomas of thyroid, different histologic patterns. (A) Spindle cell pattern. Note the coexistence of well-differentiated thyroid carcinoma (left side) (hematoxylin and eosin, ×400). (B) Giant cell pattern (hematoxylin and eosin, ×300). (C) Epidermoid pattern (hematoxylin and eosin, ×300).

is a relatively new distinct subtype of thyroid carcinoma, and the criteria for diagnosis is not uniform among pathologists. For instance, only 3 (14%) tumors were labeled originally by pathologists as insular carcinomas, and they were diagnosed in recent years. In this study, by reviewing the histologic features of all the thyroid carcinomas managed in our institution for the past 45 years and applying strict histologic criteria, the incidence of insular carcinoma was found to be 3% of the primary thyroid carcinomas.

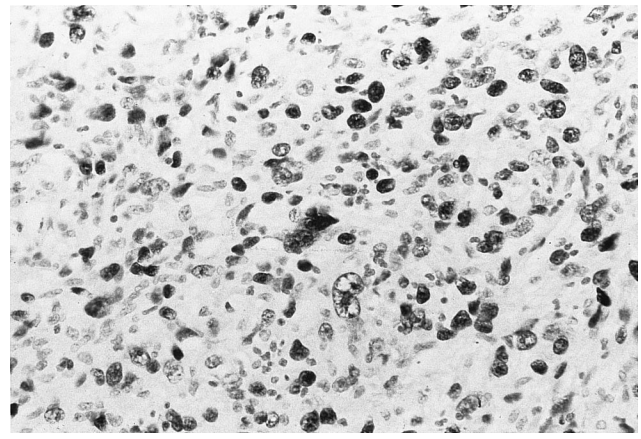

Figure 4. p53 expression in anaplastic carcinoma of the thyroid (×400).

Concomitant well-differentiated thyroid carcinomas were noted in both anaplastic and insular carcinomas. Foci of insular carcinoma were also noted in the anaplastic carcinomas. These findings strengthen the postulation that well-differentiated carcinoma can progress to insular carcinoma and then to anaplastic carcinoma of the thyroid by dedifferentiation.[20,21] Also, long-standing goiter, present in approximately 25% of patients with either anaplastic or insular carcinoma, contributes to the development of these aggressive subtypes of thyroid carcinomas.

Calcification with or without osseous metaplasia was seen in the stroma of 23% of insular carcinomas and 47% of anaplastic carcinomas. The high incidence of calcification or ossification was unusual and had not been reported in other series. Although calcification or ossification could take place in a few weeks, its presence might still imply that the diseases were of long standing.

Patients with insular carcinoma of the thyroid, when compared with those with anaplastic carcinoma, were younger (mean age 54 vs. 70 years, $P = .0001$) and had smaller tumors (mean diameter 5 vs. 8 cm, $P = .003$). Both

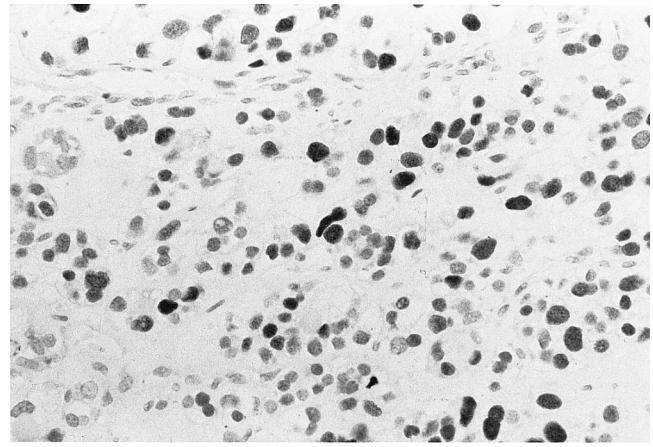

Figure 5. p21 expression in anaplastic carcinoma of the thyroid (×400).

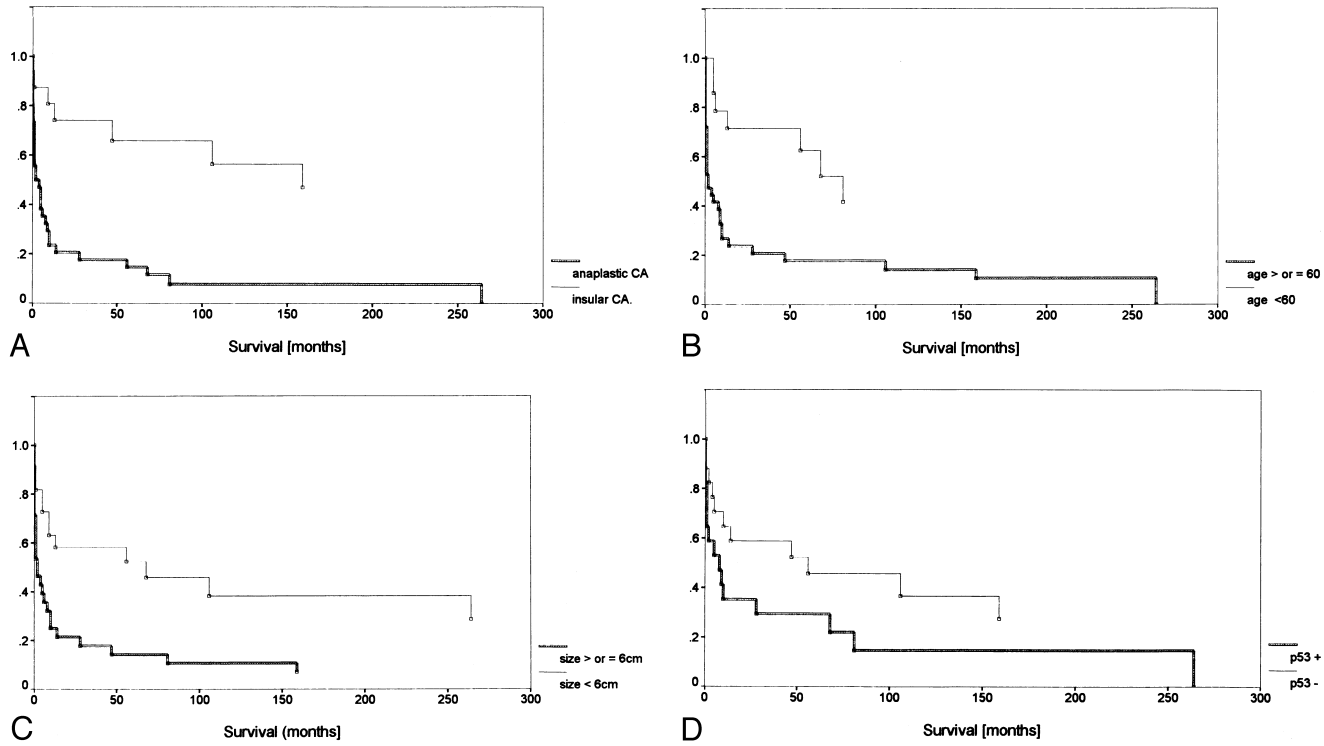

Figure 6. (**A**) Kaplan-Meier disease-specific survival with reference to histologic subtypes; $P = .00001$. (B) Kaplan-Meier disease-specific survival with respect to different age groups in patients with either anaplastic or insular carcinoma; $P = .002$. (C) Kaplan-Meier disease-specific survival with respect to tumor size in patients with either anaplastic or insular carcinoma; $P = .003$. (D) Kaplan-Meier disease-specific survival with respect to p53 overexpression in patients with either anaplastic or insular carcinoma; $P = .01$.

groups of patients often had neck masses. Patients with insular carcinomas had tumor in either lobe of the thyroid, whereas anaplastic carcinomas might involve both lobes. In anaplastic carcinoma, a significant proportion of the patients had an acute deterioration in general condition and died shortly after admission.[42]

In the current series, distant metastases were noted in only approximately one third of patients with either insular or anaplastic carcinomas, despite the high death rates. This indicates the importance of local disease. According to the information gathered from the literature and in the current series, distant metastases in insular carcinoma of the thyroid were often noted in the lungs and bones.[1–8] In our patients, metastases were also noted in the liver and heart. In anaplastic carcinomas, distant metastases were noted in a variety of organs. In the current series, the three most common sites of metastases in anaplastic carcinoma of the thyroid were lung, bone, and heart. Other sites of distant metastases included kidney, adrenal gland, brain, pancreas, and salivary gland.

It has been difficult to draw definite conclusions on the behavior of insular carcinoma of the thyroid. This is partly because of the short and incomplete follow-up in many studies. The current study, compared with the other studies on insular carcinomas, had the longest mean follow-up period (10 years). In our previous studies, the 5- and 10-year survival rates for patients with follicular carcinoma were 87% and 80%, respectively,[43] whereas during a median follow-up of 10 years, the survival rate for patients with papillary carcinoma was 92%.[44] In the present study, the 5- and 10-year survival rates for patients with insular carcinoma were 46% and 42%, respectively; those for anaplastic carcinomas were 15% and 3%, respectively. Thus, we can conclude that insular carcinoma has an intermediate position between well-differentiated carcinoma and anaplastic carcinoma with regard to both histologic features and biologic aggressiveness.

Anaplastic carcinoma of the thyroid remains one of the most lethal tumors because of its advanced presentation in elderly patients in poor general condition, the difficulty in diagnosis, and the lack of effective treatment.[42] In our unit, surgery was adopted as the first-line treatment—debulking the tumor and ensuring an airway—followed by postoperative combination chemotherapy and radiotherapy in selected patients. Insular carcinoma of the thyroid had a better prognosis, and aggressive treatment is beneficial to the patients. Nevertheless, the tumor is uncommon and rarely diagnosed correctly before thyroidectomy. The current protocol of management is total thyroidectomy followed by adjuvant treatment (radioactive iodide) and close follow-up.

Genetic alternations in Ret, p53, and Ras are important in the pathogenesis of many thyroid carcinomas. Rearrangement of Ret is found in many papillary carcinomas of the thyroid.[45] p53 mutation has been proposed to have a role in

Table 3. SUMMARY OF STUDIES OF THE RELATION BETWEEN p53 ALTERATIONS AND THYROID CARCINOMAS (ANAPLASTIC AND POORLY DIFFERENTIATED)

Author/Year/Place	% Positive (no. positive/no. tested)		Methods Used	Remarks
	Poorly Differentiated Carcinoma	Anaplastic Carcinoma		
Wyllie/1989/UK[13]	—	0% (0/4)	Southern, Northern blots	
Wright/1991/UK, Germany[14]	—	0% (0/20)	IM, sequencing	
Nakamura/1992/Japan[15]	—	22% (2/9)	PCR-RNAase protection analysis	
Yoshimoto/1992/Japan[16]	—	0% (0/1)	PCR, SSCP	
Dobashi/1993/Japan[18]	41% (9/22)	64% (7/11)	IM	
[Dobashi/1994/Japan][17]	33% (2/6)	66% (4/6)	PCR, SSCP, sequencing	Done on IM-positive cases
Fagin/1993/USA[19]	—	83% (5/6)	PCR, SSCP	
Ito/1993/Japan[20]	—	87% (7/8)	PCR-RFLP, sequencing	
[Ito/1992/Japan][21]	—	86% (6/7)	PCR, sequencing	
Zou/1993/Saudi Arabia[22]	—	20% (1/5)	RT-PCR, SSCP	
Pilotti/1994/Italy[25]	57% (8/14)	55% (12/22)	IM	
[Donghi/1993/Italy][24]	25% (2/8)	71% (5/7)	IM, PCR, SSCP	
[Pilotti/1994/Italy][23]	—	100% (5/5)	IM	Done on cases with dedifferentiated carcinoma
Soares/1994/Spain[26]	16% (5/31)	83% (10/12)	IM	
Holm/1994/Norway[27]	—	75% (18/24)	IM	
Matias-Guiu/1994/Spain[28]	—	50% (2/4)	IM	Cases arising from papillary carcinomas
Cameselle-Teijeiro/1995/Spain[29]	—	0% (0/1)	IM	A case of papillary and mucoepidermoid carcinoma with anaplastic transformation
Yane/1996/Japan[30]	—	50% (1/2)	IM, PCR, SSCP, sequencing	
Salvatore/1996/Italy[31]	0% (0/2)	25% (1/4)	IM, PCR, SSCP, sequencing	
Pollina/1996/Italy[32]	5% (1/20)	63% (15/24)	IM	
[Basolo/1997/Italy][33]	9% (1/11)	71% (15/21)	IM	
Matias-Guiu/1996/Spain[34]	—	100% (1/1)	IM, PCR, SSCP, sequencing	A case of follicular carcinoma with foci of poorly differentiated and anaplastic carcinoma
Ho/1996/Taiwan[35]	17% (5/29)	0% (0/4)	IM, PCR, SSCP, sequencing	1 anaplastic carcinoma was positive by IM
Jossart/1996/USA[36]	—	0% (0/2)	PCR-DGGE	
Zedenius/1996/Sweden[37]	—	25% (1/4)	IM, PCR, CDGE, sequencing	All 4 were positive for IM
Ito/1996/Japan[38]	—	53% (10/19)	IM	
Moore/1998/Canada[39]	100% (1/1)	100% (2/2)	IM	
Present study/1999/Hong Kong	0% (0/13)	69% (20/29)	IM	

[ ]: studies reported by the same group of authors, probably reporting similar but smaller number of cases.
CDGE, constant denaturing gel electrophoresis; DGGE, denaturing gradient gel electrophoresis; IM, immunohistochemistry; PCR; polymerase chain reaction; RFLP, restriction fragment length polymorphism; RT, reverse transcription; SSCP; single strand conformation polymorphism.

the stepwise dedifferentiation of human thyroid carcinomas, from well-differentiated thyroid carcinoma to poorly differentiated thyroid carcinoma to anaplastic carcinoma.[17] Also, mutations in the Ras oncogene were noted in insular carcinoma and follicular carcinoma of the thyroid.[7] In the present study, well-differentiated thyroid carcinomas and poorly differentiated (insular) thyroid carcinomas showed no p53 overexpression. In contrast, 69% of anaplastic carcinomas had overexpression of p53. Thus, p53 overexpression seems to have a role in anaplastic carcinoma but not in the other, less aggressive thyroid carcinomas.

Reports on the incidence of p53 alterations in poorly differentiated and anaplastic carcinomas of the thyroid are shown in Table 3. Summarizing the findings in the literature, approximately half of anaplastic carcinomas revealed p53 alterations.[13–39] p53 alterations in poorly differentiated carcinomas of the thyroid were difficult to analyze because many studies did not clearly define what was meant by the

Table 4. SUMMARY OF STUDIES ON THE RELATION BETWEEN p21 ALTERATIONS AND THYROID CARCINOMAS (ANAPLASTIC AND POORLY DIFFERENTIATED)

Author/Year/Place	% Positive (number positive/number tested)		Methods Used	Remarks
	Poorly Differentiated Carcinoma	Anaplastic Carcinoma		
Zedenius/1996/Sweden[37]	—	100% (4/4)	Immunohistochemistry	All p21-positive cases were also positive for p53 overexpression.
Ito/1996/Japan[38]	—	26% (5/19)	Immunohistochemistry	4 of the 5 p21-positive cases were also positive for p53 overexpression.
Shi/1996/Saudi Arabia[40]	—	0% (0/5)	RT-PCR, SSCP	
Present study	0% (0/13)	3% (1/29)	Immunohistochemistry	The p21-positive case was also positive for p53 overexpression.

PCR, polymerase chain reaction; RT, reverse transcription; SSCP, single strand conformation polymorphism.

term "poorly differentiated thyroid carcinoma." Nevertheless, in the literature, slightly more than 20% of poorly differentiated thyroid carcinomas showed p53 alterations. Thus, anaplastic carcinomas showed a higher overall incidence of p53 alterations than poorly differentiated thyroid carcinomas. In this study, when we applied strict histologic criteria for poorly differentiated thyroid carcinoma, we found no evidence of p53 overexpression.

There were no previous studies documenting the level of p21 expression in insular carcinoma, and there were only a few studies on the role of p21 in anaplastic carcinoma of the thyroid (Table 4).[37,38,40] Shi et al[40] found no mutation of p21 in five anaplastic carcinomas of the thyroid. Zedenius et al[37] and Ito et al[38] found p21 expression in 4 of 4 patients (100%) and 5 of 19 patients (26%) with anaplastic carcinoma of the thyroid, respectively. In the current study, with more patients, p21 expression was noted in 3% of the 29 anaplastic carcinomas and was absent in 13 insular carcinomas. Thus, there seems to be a loss of p21 expression in both insular and anaplastic carcinoma of the thyroid.

The degree of p21 immunoreactivity is assumed to reflect the degree of expression of the normal p21 gene. In theory, p53 expression should correlate with a lack of p21 expression, and vice versa. This association was either confirmed or disproved in some studies.[46,47] In anaplastic carcinomas of the thyroid (both in the literature and in our series), nearly all the p21-positive tumors were also positive for p53. The results indicated that the expression of p21 could be induced by a p53-independent pathway. The molecular mechanisms of carcinogenesis in this tumor appear to be complicated.

In conclusion, our study showed that anaplastic carcinoma and insular carcinoma, although uncommon, are important subtypes of thyroid carcinoma with distinct clinicopathologic features. p53 overexpression was noted in anaplastic carcinoma but not in insular carcinoma, suggesting that p53 overexpression might be important in the pathogenesis of the former.

References

1. Carcangiu ML, Zampi G, Rosai J. Poorly differentiated ("insular") thyroid carcinoma. A reinterpretation of Langhans' "wuchernde Struma." Am J Surg Pathol 1984; 8:655–668.
2. Flynn SD, Forman BH, Stewart AF, Kinder BK. Poorly differentiated ("insular") carcinoma of the thyroid gland: an aggressive subset of differentiated thyroid neoplasms. Surgery 1988; 104:963–970.
3. Killeen RM, Barnes L, Watson CG, et al. Poorly differentiated ("insular") thyroid carcinoma. Report of two cases and review of the literature. Arch Otolaryngol Head Neck Surg 1990; 116:1082–1086.
4. Justin EP, Seabold JE, Robinson RA, et al. Insular carcinoma: a distinct thyroid carcinoma with associated iodine-131 localization. J Nucl Med 1991; 32:1358–63.
5. Papotti M, Botto-Micca F, Favero A, et al. Poorly differentiated thyroid carcinomas with primordial cell component: a group of aggressive lesions sharing insular, trabecular, and solid patterns. Am J Surg Pathol 1993; 17:291–301.
6. Sasaki A, Daa T, Kashima K, et al. Insular component as a risk factor of thyroid carcinoma. Pathol Int 1996; 46:939–946.
7. Pilotti S, Collini P, Mariani L, et al. Insular carcinoma: a distinct de novo entity among follicular carcinomas of the thyroid gland. Am J Surg Pathol 1997; 21:1466–1473.
8. Albareda M, Puig-Domingo M, Wengrowicz S, et al. Clinical forms of presentation and evolution of diffuse sclerosing variant of papillary carcinoma and insular variant of follicular carcinoma of the thyroid. Thyroid 1998; 8:385–391.
9. Sakamoto A, Kasai N, Sugano H. Poorly differentiated carcinoma of the thyroid. A clinicopathologic entity for a high-risk group of papillary and follicular carcinomas. Cancer 1983; 52:1849–1855.
10. Hollstein M, Sidransky D, Vogelstein B, Harris CC. p53 mutations in human cancers. Science 1991; 253:49–53.
11. El-Deiry WS, Tokino T, Velculescu VE, et al. WAF1, a potential mediator of p53 tumor suppression. Cell 1993; 75:817–825.
12. Michieli P, Chedid M, Lin D, et al. Induction of WAF1/CIP1 by a p53-independent pathway. Cancer Res 1994; 54:3391–3395.
13. Wyllie FS, Lemoine NR, Williams ED, Wynford-Thomas D. Structure and expression of nuclear oncogenes in multi-stage thyroid tumorigenesis. Br J Cancer 1989; 60:561–565.
14. Wright PA, Lemoine NR, Goretzki PE, et al. Mutation of the p53 gene in a differentiated human thyroid carcinoma cell line, but not in primary thyroid tumours. Oncogene 1991; 6:1693–1697.

15. Nakamura T, Yana I, Kobayashi T, et al. p53 gene mutations associated with anaplastic transformation of human thyroid carcinomas. Jpn J Cancer Res 1992; 83:1293–1298.
16. Yoshimoto K, Iwahana H, Fukuda A, et al. Role of p53 mutations in endocrine tumorigenesis: mutation detection by polymerase chain reaction—single strand conformation polymorphism. Cancer Res 1992; 52:5061–5064.
17. Dobashi Y, Sugimura H, Sakamoto A, et al. Stepwise participation of p53 gene mutation during dedifferentiation of human thyroid carcinomas. Diagn Mol Pathol 1994; 3:9–14.
18. Dobashi Y, Sakamoto A, Sugimura H, et al. Overexpression of p53 as a possible prognostic factor in human thyroid carcinoma. Am J Surg Pathol 1993; 17:375–381.
19. Fagin JA, Matsuo K, Karmakar A, et al. High prevalence of mutations of the p53 gene in poorly differentiated human thyroid carcinomas. J Clin Invest 1993; 91:179–184.
20. Ito T, Seyama T, Mizuno T, et al. Genetic alterations in thyroid tumor progression: association with p53 gene mutations. Jpn J Cancer Res 1993; 84:526–531.
21. Ito T, Seyama T, Mizuno T, et al. Unique association of p53 mutations with undifferentiated but not with differentiated carcinomas of the thyroid gland. Cancer Res 1992; 52:1369–1371.
22. Zou M, Shi Y, Farid NR. p53 mutations in all stages of thyroid carcinomas. J Clin Endocrinol Metab 1993; 77:1054–1058.
23. Pilotti S, Collini P, Del-Bo R, et al. A novel panel of antibodies that segregates immunocytochemically poorly differentiated carcinoma from undifferentiated carcinoma of the thyroid gland. Am J Surg Pathol 1994; 18:1054–1064.
24. Donghi R, Longoni A, Pilotti S, et al. Gene p53 mutations are restricted to poorly differentiated and undifferentiated carcinomas of the thyroid gland. J Clin Invest 1993; 91:1753–1760.
25. Pilotti S, Collini P, Rilke F, et al. Bcl-2 protein expression in carcinomas originating from the follicular epithelium of the thyroid gland. J Pathol 1994; 172:337–342.
26. Soares P, Cameselle-Teijeiro J, Sobrinho-Simoes M. Immunohistochemical detection of p53 in differentiated, poorly differentiated and undifferentiated carcinomas of the thyroid. Histopathology 1994; 24: 205–210.
27. Holm R, Nesland JM. Retinoblastoma and p53 tumour suppressor gene protein expression in carcinomas of the thyroid gland. J Pathol 1994; 172:267–272.
28. Matias-Guiu X, Cuatrecasas M, Musulen E, Prat J. p53 expression in anaplastic carcinomas arising from thyroid papillary carcinomas. J Clin Pathol 1994; 47:337–339.
29. Cameselle-Teijeiro J, Febles-Perez C, Sobrinho-Simoes M. Papillary and mucoepidermoid carcinoma of the thyroid with anaplastic transformation: a case report with histologic and immunohistochemical findings that support a provocative histogenetic hypothesis. Pathol Res Pract 1995; 191:1214–1221.
30. Yane K, Konishi N, Kitahori Y, et al. Lack of p16/CDKN2 alterations in thyroid carcinomas. Cancer Lett 1996; 101:85–92.
31. Salvatore D, Celetti A, Fabien N, et al. Low frequency of p53 mutations in human thyroid tumours; p53 and Ras mutation in two out of fifty-six thyroid tumours. Eur J Endocrinol 1996; 134:177–183.
32. Pollina L, Pacini F, Fontanini G, et al. bcl-2, p53 and proliferating cell nuclear antigen expression is related to the degree of differentiation in thyroid carcinomas. Br J Cancer 1996; 73:139–143.
33. Basolo F, Pollina L, Fontanini G, et al. Apoptosis and proliferation in thyroid carcinoma: correlation with bcl-2 and p53 protein expression. Br J Cancer 1997; 75:537–541.
34. Matias-Guiu X, Villanueva A, Cuatrecasas M, et al. p53 in a thyroid follicular carcinoma with foci of poorly differentiated and anaplastic carcinoma. Pathol Res Pract 1996; 192:1242–1249.
35. Ho YS, Tseng SC, Chin TY, et al. p53 gene mutation in thyroid carcinoma. Cancer Lett 1996; 103:57–63.
36. Jossart GH, Epstein HD, Shaver JK, et al. Immunocytochemical detection of p53 in human thyroid carcinomas is associated with mutation and immortalization of cell lines. J Clin Endocrinol Metab 1996; 81:3498–3504.
37. Zedenius J, Larsson C, Wallin G, et al. Alterations of p53 and expression of WAF1/p21 in human thyroid tumors. Thyroid 1996; 6:1–9.
38. Ito Y, Kobayashi T, Takeda T, et al. Expression of p21 (WAF1/CIP1) protein in clinical thyroid tissues. Br J Cancer 1996; 74:1269–74.
39. Moore D, Ohene-Fianko D, Garcia B, Chakrabarti S. Apoptosis in thyroid neoplasms: relationship with p53 and bcl-2 expression. Histopathology 1998; 32:35–42.
40. Shi Y, Zou M, Farid NR, al-Sedairy ST. Evidence of gene deletion of p21 (WAF1/CIP1), a cyclin-dependent protein kinase inhibitor, in thyroid carcinomas. Br J Cancer 1996; 74:1336–1341.
41. Rosai J, Carcangiu ML, Delellis RA. Undifferentiated (anaplastic carcinoma). In: Rosai J, Sobin LH, ed. Atlas of Tumor Pathology: Tumors of the Thyroid Gland. Washington DC: AFIP third series, fascicle 5; 1992:135–159.
42. Lo CY, Lam KY, Wan KY. Anaplastic carcinoma of the thyroid. Am J Surg 1999; 177:337–339.
43. Lo CY, Lorentz TG, Wan KY. Follicular carcinoma of the thyroid gland in Hong Kong Chinese. Br J Surg 1993; 82:1095–1097.
44. Lorentz TG, Lau PWK, Lo CY, et al. Multivariate analysis of risk factors influencing survival in 110 ethnic Chinese with papillary thyroid cancer. World J Surg 1994; 18:547–551.
45. Lam AKY, Montone KT, Nolan KA, LiVolsi VA. Ret oncogene activation in papillary thyroid carcinoma: prevalence and implication on the histological parameters. Hum Pathol 1998; 29:565–568.
46. Backe J, Gassel AM, Hauber K, et al. p53 protein in endometrial cancer is related to proliferative activity and prognosis but not to expression of p21 protein. Int J Gynecol Pathol 1997; 16:361–368.
47. Ito K, Sasano H, Matsunaga G, et al. Correlations between p21 expression and clinicopathological findings, p53 gene and protein alterations, and survival in patients with endometrial carcinoma. J Pathol 1997; 183:318–324.